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Environmental sampling – A tool to verify the effectiveness of preventive hygiene measures*

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The environment in which foods are prepared and handled can have significant impact on microbiological safety and quality, particularly for ready-to-eat foods. Outbreaks associated with *Listeria monocytogenes* and salmonellae have led to an increasing awareness that certain pathogens can persist in food processing environments, contaminate foods as they are being processed and lead to foodborne illness (1, 2). To prevent such unacceptable contamination many food processors have established environmental sampling programs to monitor and verify whether their Good Hygienic Practices (GHP) are effective and being correctly applied (2).

Environmental sampling programs differ in many respects from the lot acceptance sampling plans described by Drs Cordier and Dahms (see their presentations in this journal). Environmental sampling is designed to assess the effectiveness of GHP, control certain pathogens and/or spoilage microorganisms within the environment and prevent contamination of food. Thus, such programs are used as a tool to prevent contamination rather than evaluate whether food is acceptable. This agrees with the intent of GHP and HACCP; namely, to prevent food safety problems from occurring and not rely on end product testing to sort "good food" from "bad food". Another difference is that the number and location of the sites selected for sampling are based on experience with the specific food operation and are not statistically designed. Instead, environmental sampling should be viewed as an ongoing investigational sampling program. If lot acceptance testing indicates a specific lot of food is not in compliance with specifications, environmental sampling is commonly used as the framework to investigate how the food became contaminated and how to prevent future unacceptable lots.

When establishing a routine environmental sampling program it is necessary to decide how, when and where samples will be collected. The sampling method will vary depending on the food process. Basically, the method of sampling (e.g., sponge sampling of a product contact surface, floor sweepings, residue on filters) should be

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capable of detecting unacceptable contamination. Sampling the surfaces of equipment after cleaning and disinfecting before start of operation has been common practice to verify the effectiveness of cleaning and disinfection procedures. Typically, such samples show a high level of compliance (e.g., >90 %). Sampling clean equipment, however, can lead to a false sense of security. Sampling equipment during or at the end of production is much more realistic and can reveal contamination that would otherwise go undetected.

The location of sites in the sampling program should be based on previous experience. For ready-to-eat foods a common sample site is the surface of equipment that the food contacts just before final packaging. Other sites located upstream in the process also are commonly included. The goal of the sampling program is to detect a problem, if one occurs, and prevent consumers from being exposed to an unacceptable level of pathogens. To accomplish this goal the environmental sampling program must be sufficiently strong in terms of the number of samples; how, when and where they are collected; how they are handled between sampling and analysis and the sensitivity of the analytical method. One limiting factor that manufacturers must address is the cost of the environmental sampling program. Important factors influencing cost include the number of samples, the cost of collecting and transporting the samples to the laboratory, the analytical cost and maintaining a database that can be used to detect trends and inform facility management.

Consideration should be given to sampling the food at different steps in the process as an alternative to sampling the environment. Knowledge of the impact that each step in the process has on the microbial content of the food should influence the locations where food samples should be collected. The type of food (e.g., milk, cheese, dry milk powder, cooked meat) and method of manufacture also will influence how the foods are prepared for analysis in the laboratory. For example, cooked meat products should have low internal numbers of viable bacteria and higher surface numbers due to contamination that can occur between cooking and packaging. Experience also indicates that sampling the food occasionally can detect contamination that is missed by the routine environmental sampling program. The reason for this discrepancy is not always apparent but such experiences suggest that sampling the food at some frequency should be included to supplement the environmental samples and verify the environmental program is sufficiently sensitive and functioning as expected.

Transient and resident microflora

When interpreting results from environmental samples it is important to differentiate between transient and resident microflora. Transient microorganisms can be introduced into the food environment by many routes and routine application of GHP is adequate to remove or kill them during cleaning and disinfection. In some cases, however, certain transients can become established, multiply and persist over time. Persistent microflora can exist in biofilms or in niches. Biofilms occur when

microorganisms become attached or immobilized on a surface, often within a matrix of microbially produced organic polymers, and provide favorable conditions for growth and survival (e.g., increased resistance to disinfectants). Biofilms, in general, are more common in closed systems such as in pipes, heat exchangers and on gaskets that are infrequently or inadequately cleaned. The persistence of *L. monocytogenes* in floor drains is likely due to biofilm formation. Niches occur in sites where food and moisture accumulate (e.g., inside the hollow supports and legs of equipment, inside hollow rollers for conveyors, in oil and grease reservoirs with worn seals, between close fitting materials such as metal-to-metal or metal-to-plastic units. Niches are sites that are not normally cleaned and disinfected.

Biofilms and niches are of greatest concern when located after a kill step (e.g., cooking) in a process. In both cases the environment appears visually clean and will pass inspection. Traditional sampling for indicators before start of operation to verify the equipment is clean will not detect the presence of a biofilm or niche. During production movement or vibrations of the equipment and/or flow of food through the system causes some of the microorganisms in biofilms and niches to become dislodged and contaminate the food. It is only through microbiological sampling of the equipment or food during production that biofilms and niches are revealed and corrective actions can be taken.

Factors (e.g., temperature, a_w, pH, nutrient content) that influence the ability of microorganisms to multiply in laboratory media also influence the number and type of microorganisms that occur in biofilms and niches in food processing environments. For example, a packaging room that operates at <10°C is an unlikely location for salmonellae to become established and multiply, however, *Listeria monocytogenes* would be quite capable of becoming a resident in such an environment. Rooms that are typically dry (e.g., rooms for drying sausages, blending and packaging dry ingredients, flour milling) are generally more controllable due to the lack of free moisture. Unfortunately, it is necessary to periodically use water in some dry environments to clean certain equipment to remove accumulated product residue that can lead to process inefficiencies or residue that becomes noticeable in the product. Another reason is to prevent contamination of a subsequent lot of food with an undeclared allergen. Leaking roofs, pipes and hose stations and condensation that forms when warm moist air contacts cold surfaces are additional sources of moisture that can occur.

Two factors influence the effectiveness of an environmental sampling program. They are 1) the ability of the sampling program to detect a problem and 2) the response by facility management when a problem is detected.

Strategies for design and implementation of a sampling program

Several strategies have been found to be effective for controlling undesirable microorganisms in food processing environments.

The first and best strategy is to prevent conditions that lead to biofilms and niches. This strategy may involve redesign of equipment using materials that are durable, have smooth surfaces and are cleanable. Design engineers must become aware of the impact they can have on pathogen control and ensuring food safety. The preventive maintenance program should include scheduled replacement or repair of equipment before it becomes a source of contamination. Equipment should be inspected periodically for parts that are cracked, worn or have developed spaces where food and moisture accumulate. The regimen for cleaning and disinfecting should include sites known to form biofilms that can lead to contamination of the food being processed.

The second strategy is to establish an effective environmental sampling program. The purpose of the sampling program is to detect in a timely manner unacceptable microbiological contamination. A positive result should be viewed as a success rather than a failure. The worst possible scenario is one in which a sampling program is incapable of detecting contamination and facility management believes the environment is in control.

An example of a sampling program could involve collecting samples during production on a weekly basis from established sites. The day of the week and time of sample collection are randomized to reflect different conditions that occur during production. The locations are based on experience and previous successes in detecting contamination and solving problems. The majority of samples consist of sponge samples from selected surfaces (e.g., product contact, support structures, floors) since this has been found to be an effective technique for detecting contamination. The analytical method is sensitive, provides a timely result and is reasonable in cost. The method is qualitative and yields a presence/absence result that enables management to respond quickly and minimize product contamination.

The number of samples varies with the complexity of the process and the food being produced. Sample site selection can be best resolved by answering the following question. If available funds can allow only a certain number of samples (e.g., 5, 10, 20) each week, which sites would yield the best assessment of control and detect the potential for product contamination? When a facility has a favorable record of control it may be possible to composite certain samples and reduce the analytical cost. There is considerable disagreement over whether drains should be included in an environmental sampling program and how to interpret the relationship between a positive drain sample (e.g., for *L. monocytogenes*) and the potential for product contamination.

The third strategy is to tabulate the results at frequent intervals (e.g., weekly) to provide a short term assessment of control. The report should include results for the past seven samplings so trends and patterns can be observed. Experience has demonstrated that the sampling program should be more stringent (e.g., more sample sites and/or frequent samplings) during periods when construction is occurring and when new or modified equipment has been installed.

The fourth strategy is to provide a quarterly or annual report because a longer term review of the data can reveal low level, intermittent contamination that may otherwise go unnoticed.

Response to results showing a problem

A major weakness throughout the food industry has been in the response to information that indicates a problem. This can be due to failure to organize the results in a manner that facilitates review, failure to recognize an evolving problem or its significance, simply filing results without review or, finally, an individual or group is not assigned responsibility or held accountable for identifying and responding to a problem. An inherent weakness in industry's response is the difficulty and time needed to detect the source(s) of contamination.

When striving to detect the source(s) all samples should be analyzed individually rather than as composites, samples should be collected more frequently (e.g., every four hours) and additional sites should be included. A map should be created that shows the layout of equipment in the rooms and the sites, dates and times when positive results have occurred. Do the results reveal patterns with certain equipment showing more positives? Where in the flow of food through the process do the first positives occur? In general, microorganisms flow downstream from the source of contamination with the food.

Fingerprinting isolates can be a very useful tool for identifying the source and the pathways of contamination. The presentation on cold smoked salmon by Dr. Gram in this journal provides an excellent example of how this technology can be used in problem solving. Through the process of continuous improvement the results from an environmental sampling program can be used to control targeted bacteria, ensure consumer safety, comply with product specifications and meet regulatory requirements.

Summary

There is increasing awareness that certain pathogens can persist in food processing environments, contaminate the food and lead to foodborne illness. Two pathogens are of particular concern, salmonellae and *Listeria monocytogenes*. Many examples could be cited where these pathogens have become established in the food processing environment and resulted in the contamination of multiple lots of food produced over long periods of time (weeks, months, years). As these events have occurred, large numbers of consumers have been exposed before the offending food could be identified and removed from the market.

Despite the best efforts of facility management to ensure compliance with GHP it is now evident that traditional techniques that rely on visual inspection or testing for indicators (e.g., total plate count, coliforms) to assess cleanliness are not adequate to detect persistent pathogens. This presentation describes how an environmental sampling program can be established for this purpose. The principles out-

lined also are applicable for controlling microorganisms that lead to unacceptable spoilage of food.

Zusammenfassung

Es wird zunehmend anerkannt, dass sich gewisse pathogene Keime in der Umgebung von Produktionslinien festsetzen können und in der Folge Lebensmittel kontaminieren und somit Lebensmittelvergiftungen auslösen können. Von besonderer Bedeutung sind dabei *Salmonella* und *Listeria monocytogenes*. Zahlreiche Beispiele zeigen, wie sich diese Pathogene in Produktionsstätten etablieren konnten und über längere Zeit (Wochen, Monate, Jahre) zur Kontamination zahlreicher Chargen geführt haben. Während dieser Zeit wurde eine grosse Anzahl Konsumenten den Keimen ausgesetzt, bevor die betreffenden Lebensmittel identifiziert und zurückgezogen wurden.

Es ist offensichtlich, dass trotz den grossen betrieblichen Anstrengungen die Einhaltung der GHP zu garantieren, die traditionellen Methoden zur Überprüfung der Sauberkeit, wie die visuelle Inspektion oder die Suche nach Indikatorkeimen (z.B. Gesamtkeimzahl, Coliforme), nicht ausreichen um angesiedelte Pathogene zu finden. Dieser Vortrag beschreibt wie ein spezifisches Überwachungsprogramm zu diesem Zweck eingeführt werden kann. Die diskutierten Grundprinzipien können auch zur Kontrolle von unerwünschten Mikroorganismen verwendet werden, welche zum Verderb von Lebensmittel führen können.

Résumé

Il est de plus en plus reconnu que certains pathogènes peuvent survivre dans l'environnement de lignes de production, causer leur contamination et donc provoquer des intoxications alimentaires. Deux pathogènes sont particulièrement importants, *Salmonella* et *Listeria monocytogenes*. De nombreux exemples pourraient être cités qui démontrent comment ces pathogènes peuvent s'établir dans des sites de productions et contribuer à la contamination de nombreuses charges de produits durant des périodes prolongées (semaines, mois, années). De ce fait durant cette période de nombreux consommateurs ont été exposés avant le retrait des produits.

Malgré des efforts importants pour maintenir les Bonnes Pratiques d'Hygiène dans les établissements, il apparaît aujourd'hui clairement que les méthodes traditionnelles de vérification des nettoyages comme l'inspection visuelle ou la recherche d'indicateurs (p.ex. flore aérobie, coliformes) ne sont pas suffisantes à la détection de pathogènes établis. Cet exposé décrit l'introduction d'un programme spécifique à cette fin. Les principes discutés peuvent également être appliqués afin de maîtriser des germes indésirés causant des altérations de produits.

Key words

Food, environment, sampling, *L. monocytogenes, Salmonella*

References

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